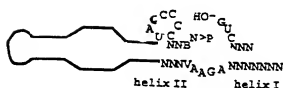


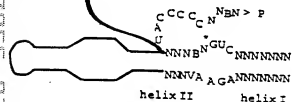
Figure 1.

Trans Cleavage and Ligation

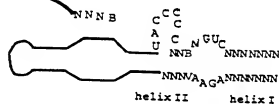
- T7 promoter Library Sequences
- Transcribe library with T7 RNA polymerase.
 - Cis-cleavage occurs.
 - Purify those randomized ribozymes that have undergone cis-cleavage.



React with target
RNA in Trans



Trans Ligation



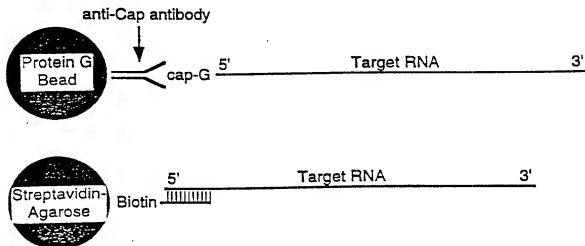
- Trans-ligated products are isolated and amplified by RT-PCR.
- Trans-ligated ribozymes can then be further amplified and subcloned into AAV vectors for production of a target specific ribozyme gene vector library.

Fig. 2

10067955-020502

Attaching RNA target to solid support

Binding target RNA at 5' end:



Binding target RNA at 3' end:

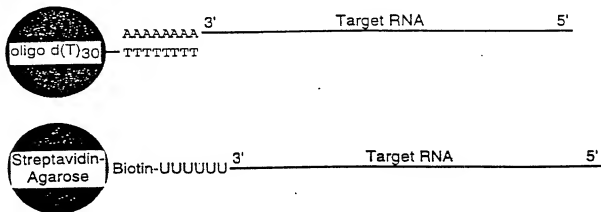


Fig. 3

In vitro selection of optimal ribozymes

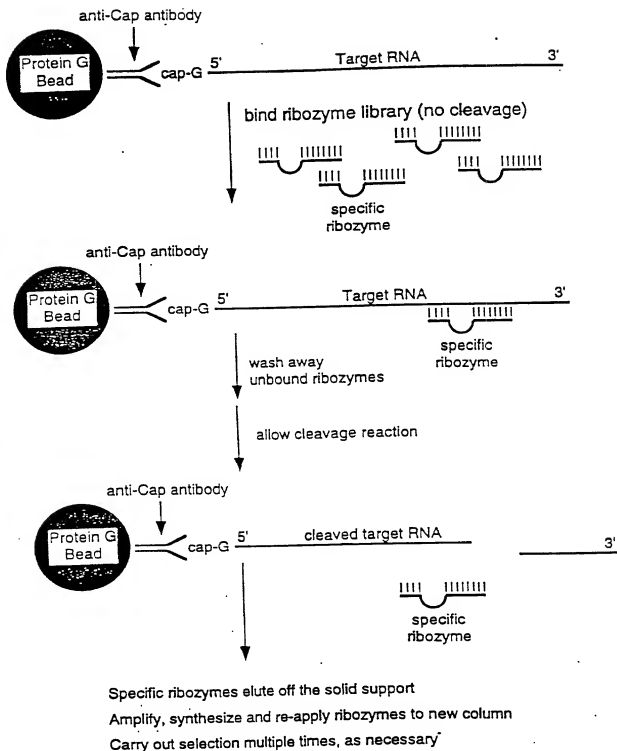


Fig. 4

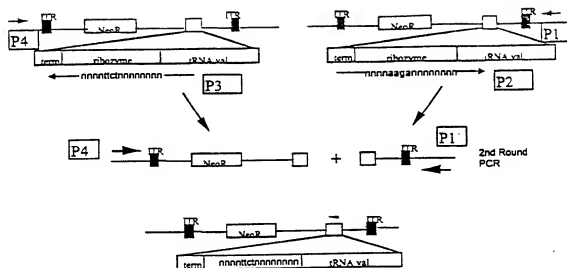


Fig. 5

Cleavage of Various Target RNAs with AAV- Ribozyme Vector Library

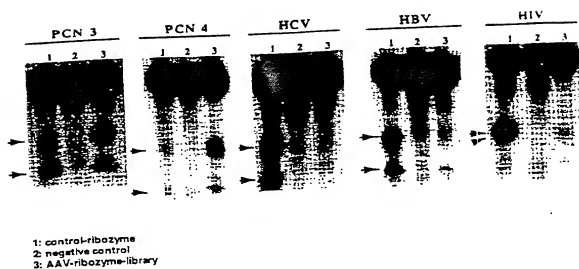


Fig. 6

p1014/2kb

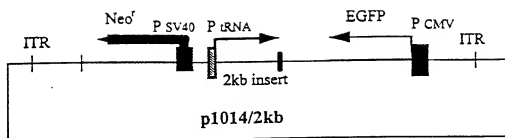


Fig. 7

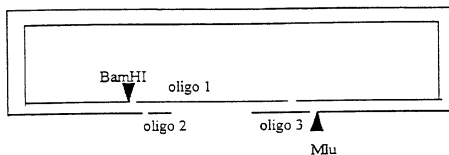


Fig. 8

205020-95679007

pAAV-Hygro-PGK

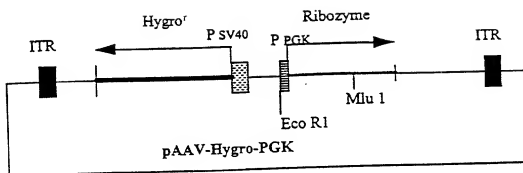


Fig. 9

pPol II/PGKmus/neo

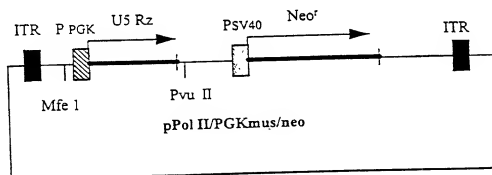


Fig. 10

10067956.020502

p 1015

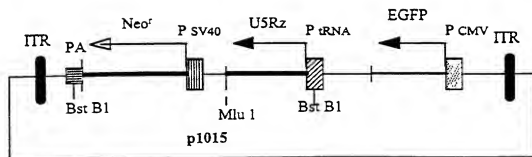
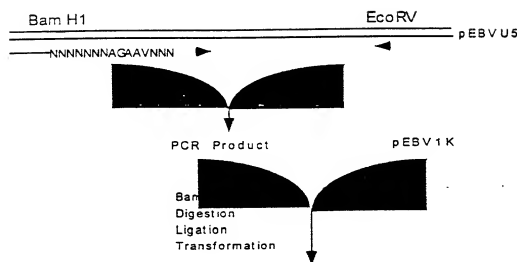


Fig. 11



ERL030398

Fig. 12

10067956.020502

pLHPM-2kb

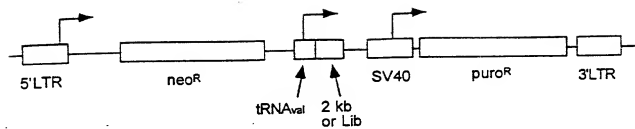


Fig. 13a

pLPR-2kb

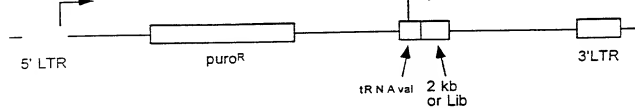


Fig. 13b

200502095679067

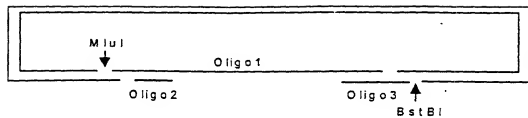


Fig. 14

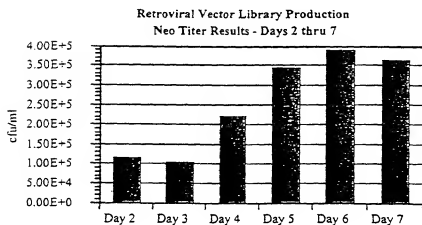


Fig. 15

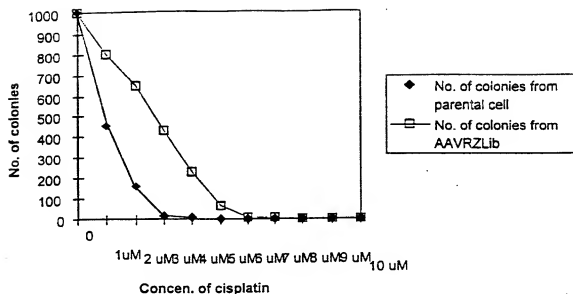


Fig. 16

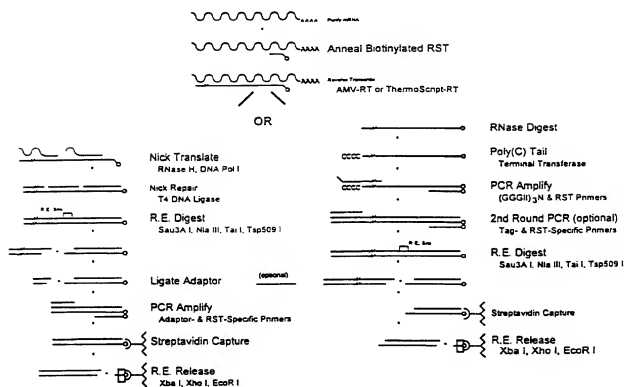


Fig. 17

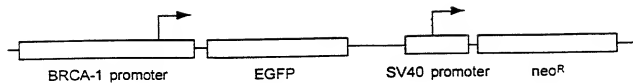


Fig. 18

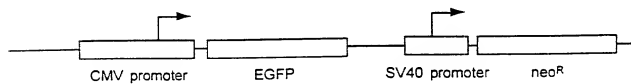


Fig. 19

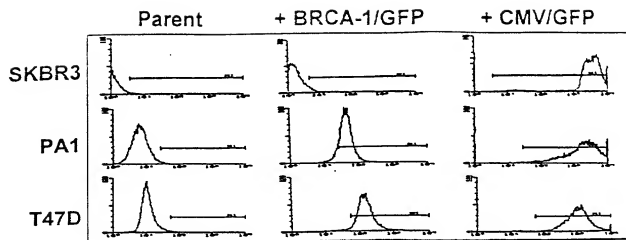


Fig. 20

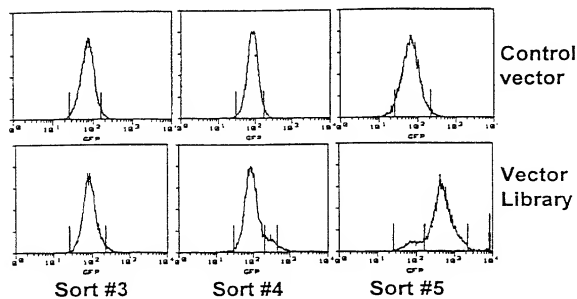


Fig. 21

SV 40 - hygro - HCV IRES - HSV-tk

Fig. 22

Transfection (Electroporation)

HeLa

hygromycin
selection

stable cell line
SV40-hygro-IRES-tk (5'TK)

no ribozyme

no inhibition of
IRES translation

HSV-tk expression

cell death

RzLibrary

inhibition of
IRES translation

no HSV-tk

cell survival

Ganciclovir

Fig. 23

Figure 24

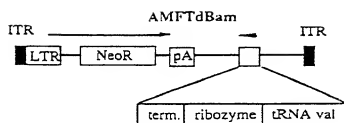


Figure 25

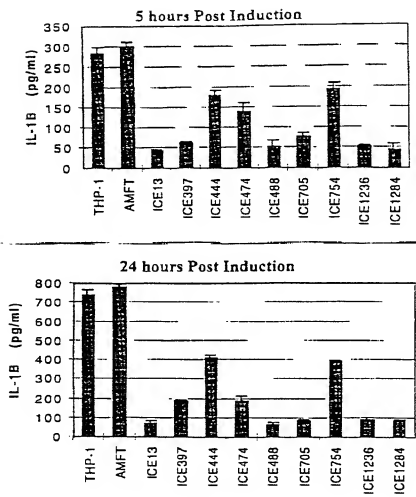
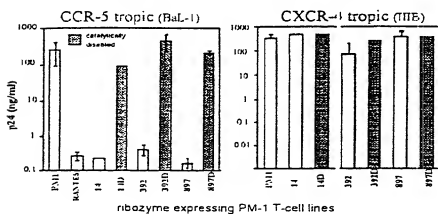


Figure 26

Resistance to CCR-5 tropic HIV



Surface expression

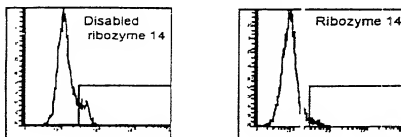
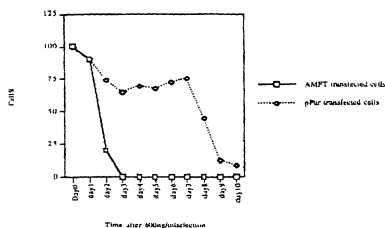


Figure 27

Puromycin selection on pPur and AMFT transfected A549 cells



Puromycin selection on pPur and AMFT transfected HeLa cells

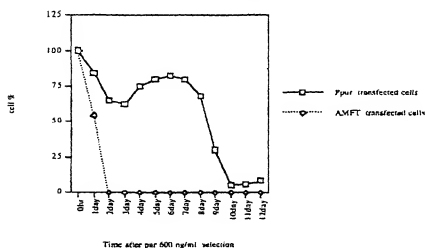
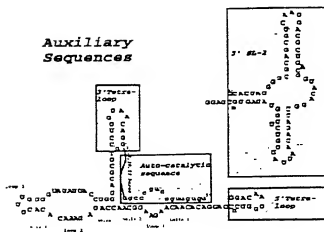


Figure 28

Auxiliary Sequences



10067956-02000000

Figure 29

Auxiliary Sequences

Time Course for Cleavage by US - SLII Chimeric Ribozymes

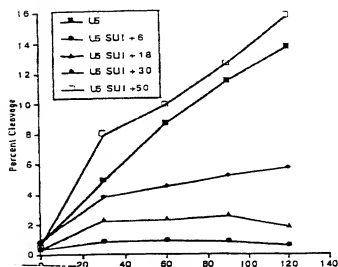
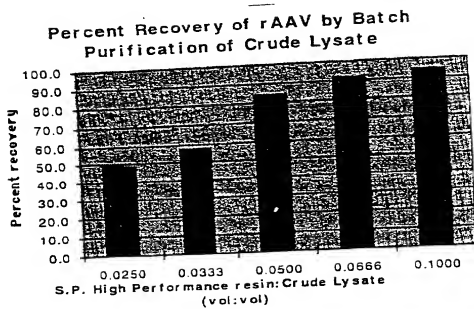


Figure 30



205020-95679001